# New records of *Sturnira koopmanhilli* (McCarthy, Albuja & Alberico, 2006) (Chiroptera, Phyllostomidae) from western and eastern Ecuador

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**Abstract.** Sturnira koopmanhilli (McCarthy, Albuja & Alberico, 2006) is a rare and vulnerable bat species distributed mainly in the Chocó biogeographic region of Ecuador and Colombia. A field expedition in northwestern Ecuador collected two new voucher specimens and tissues. We sequenced the whole *cyt-b* gene to confirm the taxonomic identity of these specimens. Furthermore, we reviewed three natural history collections to find unreported specimens. The present study provides seven new records for *S. koopmanhilli* in Ecuador and expands this species' occurrence to the eastern slopes of the Andes.

**Key words.** Chocó, cloud forests, diastemata, Neotropics, procumbent, specialized species, vulnerable species

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# **INTRODUCTION**

Sturnira koopmanhilli (McCarthy, Albuja & Alberico, 2006) (Chiroptera, Phyllostomidae) is a rare bat species distributed mainly in the Chocó biogeographic region of Ecuador and Colombia (Velazco and Patterson 2013; Solari et al. 2019). It is only known from 57 voucher specimens from 14 localities (McCarthy et al. 2006; Tirira 2008; Martínez-Arias and Solari 2013; Rodríguez-Segovia and Gavilánez-Endara 2023, 2024). Sturnira koopmanhilli has been mainly recorded in primary humid and cloud forests at altitudes between 300 and 2000 m (McCarthy et al. 2006; Martínez-Arias and Solari 2013). In Ecuador, this phyllostomid is listed as vulnerable (Tirira 2021). In the last two decades, the Choco forests of Ecuador and Colombia have been severely fragmented due to human disturbances (Sierra et al. 2002; Meyer et al. 2019), which have affected the conservation of this species.

An evolutionary study established that *S. koopmanhilli* is the sister species to *S. mordax* and probably diverged from it during the Late Pliocene (3.5 Ma) to Pleistocene (1.8 Ma) (Velazco and Patterson 2013). In that study, the molecular identification of *S. koopmanhilli* was based on the cytochrome b gene (*cyt-b*) and a regulatory non-coding region of mitochondrial DNA (mtDNA) (*D-loop*, hypervariable region HVRI section) (Velazco and Patterson 2013). Only two voucher specimens from the type locality at Los Pambiles, Esmeraldas province, have been sequenced to date, CM 112804 and CM 112812 (Carnegie Museum of Natural History) (Velazco and Patterson 2013). The Instituto Nacional de Biodiversidad (INABIO), formerly known as the Museo Ecuatoriano de Ciencias Naturales (MECN), is currently leading the creation of the National Bank of Genetic Resources of Ecuador, a biorepository dedicated to the protection and conservation of Ecuador's biodiversity. INABIO is using this biorepository for the molecular identification of rare and endangered species, such as *S. koopmanhilli*. The main objective of this paper is to present new records for *S. koopmanhilli* in western and eastern Ecuador.



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## **METHODS**

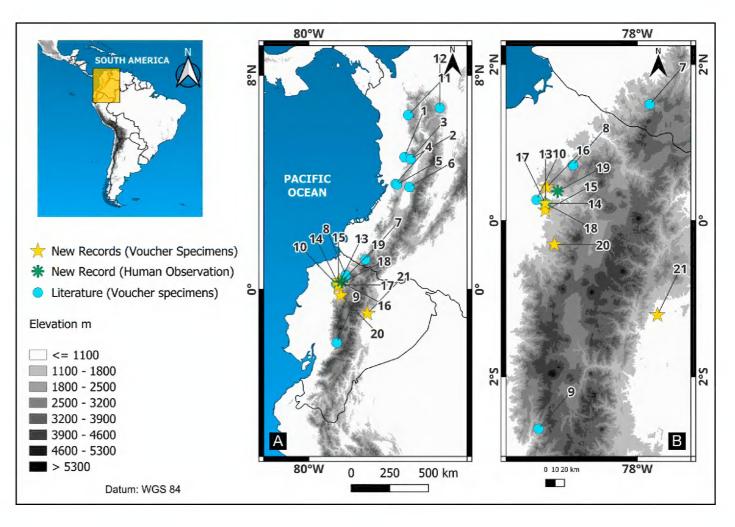
We searched for unreported or misidentified voucher specimens of *Sturnira koopmanhilli* in the mammal collections of the MECN, the Museo de la Escuela Politécnica Nacional (MEPN), and the Museo de Zoología

de la Pontificia Universidad Católica del Ecuador (QCAZ). To identify this species, we photographed the skin and skull of each collected specimen to obtain detailed information on its anatomy. We focused on the presence of elongate and procumbent incisors and the diastemata between the molars, as suggested by specialized field guides (McCarthy et al. 2006; Martínez-Arias and Solari 2013; Solari et al. 2019; Díaz et al. 2021). We used *S. koopmanhilli* MECN 6802 and MECN 6806 for comparisons (Appendix 1). In cases where it was necessary, we used Trupper digital calipers (± 0.01 mm) to obtain standard body measurements: head and body length, hind foot length, ear length, and forearm (McCarthy et al. 2006). In other cases, we obtained measurements, weight, and sex from the tag of each voucher specimen. The age and reproductive status of each specimen was determined by examining the degree of fusion of hand—wing epiphyses, nipple development, and abdominal development (Kunz and Parsons 2009). We compiled all available geographic records for this species from the literature and photographic material shared by Merlin Tuttle's Bat Conservation. This information was used to generate an updated distribution map (McCarthy et al. 2006; Martínez-Arias and Solari 2013; Rodríguez-Segovia and Gavilánez-Endara 2023, 2024) (Appendix 1).

On 16 May 2024, a field expedition was conducted to the Reserva Río Manduriacu (locality #16, Figure 1), Imbabura Province, in northwestern Ecuador. To document the bat fauna, 10 mist nets, each 12 m long, were placed in the understory. Voucher specimens were collected according to the guidelines of the American Association of Mammalogists (Sikes 2016) and the Ecuadorian Association of Mammalogy (Erazo et al. 2022). The study was carried out under collection permit no. MAATE-ARSFC-2023-0145 and an access to genetic resources permit no. MAATE-DBI-CM-2023-0334 issued by the Ministerio del Ambiente Agua y Transición Ecológica del Ecuador. Frozen tissues from two voucher specimens identified as *S. koopmanhilli* (MECN 8120 and 8121), were deposited in the mammal collection of Instituto Nacional de Biodiversidad (MECN). To verify the identity of these specimens, we extracted DNA from the liver using the GeneJET Genomic DNA Purification Kit (K0722). We amplified the entire mitochondrial cytochrome b (*cyt-b*) gene using polymerase chain reaction (PCR) with the MVZ05 forward primer, and the MVZ16 reverse primer (Smith and Patton 1993), and the GoTaq® Green Master Mix 2X kit. PCR conditions included an initial denaturation phase at 95 °C for 2 min, followed by 35 cycles of amplification. Each cycle included denaturation at 95 °C for 30 s, annealing at 45 °C for 30 s, extension at 72 °C for 80 s, and a final extension step at 72 °C for 5 min.

We sequenced the *cyt-b* gene using a MinION mk1c equipped with Flongle Flow Cells R v. 10.4.1 and the Rapid Barcoding Kit 96 (SQK-RBK114.96), following standard protocols. Dorado v. 4.3.0. was used for base calling. The resulting fastq files were filtered with a Q score of 9, and consensus sequences were generated using NGSpecies ID v. 0.3.0 (Sahlin et al. 2021). We also downloaded from GenBank, *cytb* sequences of 12 recognized *Sturnira* species (Velazco and Patterson 2013; Yánez-Fernández et al. 2023) and performed the alignment using the MAFFT algorithm in Mesquite v. 3.81 (Maddison and Maddison 2023). A maximum-likelihood tree (ML) was generated in MEGA v. 12 (Kumar et al. 2024) using the Tamura-Nei substitution model (Tamura and Nei 1993). The tree with the highest log-likelihood was selected and manually rooted by separating the outgroup branch in MEGA v. 12 (Kumar et al. 2024). Pairwise genetic distances were calculated using the same program. The *S. koopmanhilli cytb* sequences generated for this paper are available to other researchers in GenBank under accession numbers PV089741 and PV185410.

**Figure 1.** Map distribution of *Sturnira koopmanhilli* in South America. **A.** Known records in Ecuador and Colombia. **B.** Detail of the new records in Ecuador. Localities and voucher specimens are detailed in Appendix 1. The blue dot #8 corresponds to the type locality.



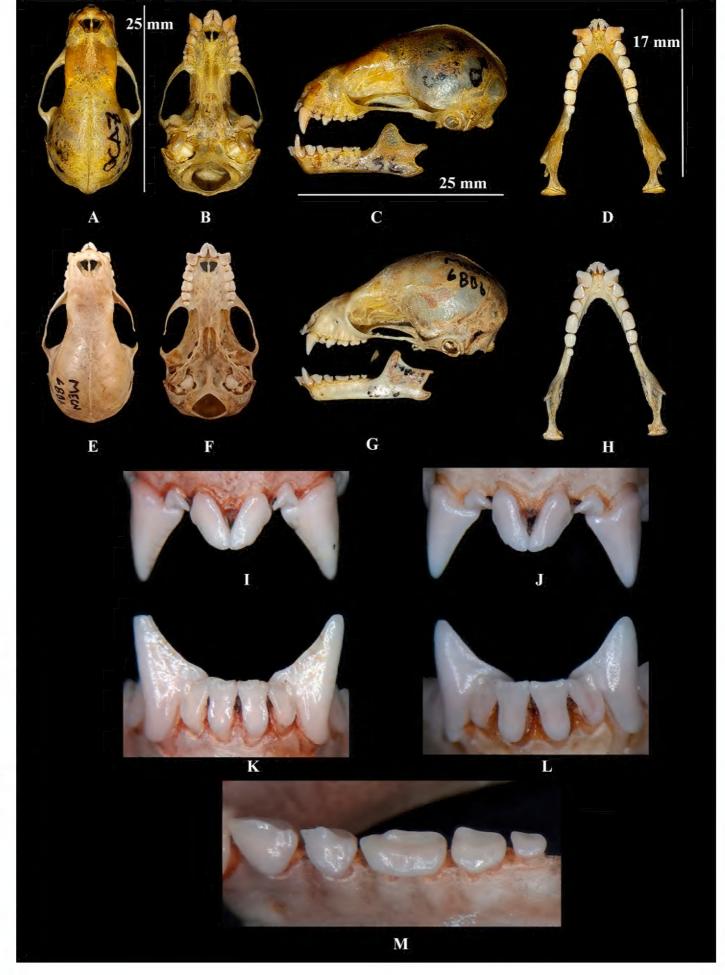


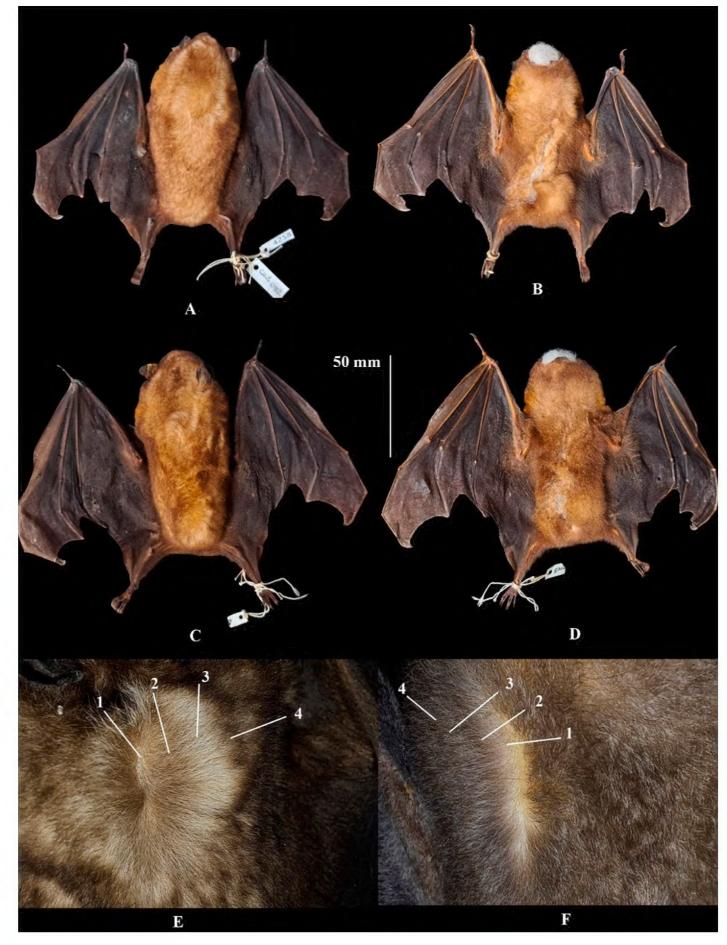
Figure 2. Skull details of Sturnira koopmanhilli. A. Dorsal view of the skull of QCAZ 4256 3. **B.** Ventral view of the skull of QCAZ 4256 3. C. Lateral view of the skull of QCAZ 4256 3. **D.** Detail of the mandible of QCAZ 4256 & (Archidona, Napo). E. Dorsal view of the skull of MECN 6806  $\mathfrak{P}$ . **F.** Ventral view of the skull of MECN 6806 ♀. **G.** Lateral view of the skull of MECN 6806 ♀. **H.** Detail of the mandible of MECN 6806 ♀ (Seven Layers Trail, Masphi Lodge, Pichincha). I. Upper inner incisors of MECN 6806 ♀. J. Upper inner incisors of MECN 6802 ♂ (Luminous Trail, Masphi Lodge, Pichincha). K. Lower incisors of MECN 6806 ♀. **L.** Lower incisors of MECN 6802 ♂. M. Lower left toothrows of MECN 6806 3. Scale bars: A–C, E–G = 25 mm; D–H = 17 mm.

# **RESULTS**

## Sturnira koopmanhilli (McCarthy, Albuja & Alberico, 2006) Figures 2–5

We found seven new geographic records of *Sturnira koopmanhilli* in Ecuador. These are based on 10 adult voucher specimens and one human observation (locality #19, Figure 1). Six records are from the Chocó biogeographical region of Ecuador. QCAZ 4256 & and QCAZ 4258 & represent the first records for this species on the eastern slopes of the Ecuadorian Andes. These specimens were collected in Archidona, Napo province (locality #21, Figure 1). Three voucher specimens of *S. koopmanhilli* deposited at the Museo de Zoología QCAZ were misidentified, corresponding to *S. ludovici* QCAZ 14738–14740 (Appendix 2).

New Records. ECUADOR — IMBABURA • Cotacachi, García Moreno, Reserva Río Manduriacu, Campamento Magnolia (locality #16, Figure 1); 00°19′10.5″N, 078°52′20.5″W; 1820 m alt.; 14 and 16 May 2024; J. Brito, F. Castellanos and L. Simba; 1♂ and 1♀, MECN 8120 and MECN 8121, ethanol-preserved specimens obtained during recent fieldwork • Bosque Protector Los Cedros, cerca de la Estación Científica los Cedros (locality #19, Figure 1); 00°16′57″N, 078°45′58″W; 1344 m alt.; 15 Apr. 2020; Daniel Whitby obs.; 1 unknown sex, photographic material shared in Merlin Tuttle's Bat Conservation website (Figure 4) — NAPO • Archidona (locality #21, Figure 1);



**Figure 3.** Skin details of *Sturnira koopmanhilli.* **A.** Dorsal view of the skin of QCAZ 4258 ♂. **B.** Ventral view of the skin of QCAZ 4258 ♂ (Archidona, Napo, Ecuador). **C.** Dorsal view of the skin of QCAZ 4256 ♂. **D.** Ventral view of the skin of QCAZ 4256 ♂ (Archidona, Napo, Ecuador). **E.** Detail of the dorsal bands of the pelage of QCAZ 18711 ♂. **F.** Detail of the ventral bands of the pelage of QCAZ 18711 ♂ (near life Center, Main Route, Mashpi Lodge, Pichincha, Ecuador). Abbreviatures: 1. Basal band. 2. Epibasal band. 3. Subdistal band. 4. Distal band. Scale bar: A−E = 50 mm.

**Figure 4**. *Sturnira koopmanhilli* at Bosque Protector Los Cedros, Imbabura, Ecuador. Courtesy of Daniel Whitby only for scientific purposes.



00°54′22.6″S, 077°48′24.5″W (estimated coordinates from QCAZ); 500–800 m alt (inferred data); 28 Dec. 2000; unknown collector; 2♂, QCAZ 4256 and QCAZ 4258, skin and skull — PICHINCHA • Quito, Pacto, San Francisco de Pachijal, Bosque maduro en el sendero que conduce a cabaña de Fundación Futuro (locality #15, Figure 1); 00°05′57.8″N, 078°53′03.2″W; 1026 m alt.; 20 Aug. 2021; Nicté Ordóñez; 1♂ and 1♀, MECN 7475 and MECN 7432, skin and skull • Quito, Pacto, Mashpi Lodge, main route, near Life Center (locality #17, Figure 1); 00°09′59.2″N, 078°53′08.7″W; 919 m alt.; 26 Aug. 2019; Joseph Cook; 1♂ and 1♀, QCAZ 18711 and QCAZ 18710, skin and skull, ethanol-preserved specimen • Quito, Pacto, Mashpi Lodge, Main Route (locality #18, Figure 1); 00°09′58.4″N, 078°53′02.8″W; 926 m alt.; 27 Aug. 2019; Joseph Cook; 1♀, QCAZ 18716, ethanol preserved specimen — Santo Domingo De Los TsáCHILAS • Santo Domingo, Alluriquín, Palmeras, Reserva Río Guajalito, vía Chiriboga-Santo Domingo (locality #20, Figure 1); 00°13′44″S, 078°47′50″W; 2200 m alt.; 30 Oct. 1991; Diego Tirira; 1♀, QCAZ 682, skin and skull.

**Identification.** We identified *S. koopmanhilli* using the following characters as suggested by McCarthy et al. (2006). The cranium is elongate and slender (Figure 2A–C, E–G). In ventral view, the posterior margin of hard palate forms a rounded or disrupted round shape (Figure 2B, F). A sulcus on the posterior surface of the upper canines is present (Figure 2B, F). The mandible has a robust coronoid and condylar process (Figure 2C, G). The maxillary and mandibular incisors are enlarged and procumbent (Figure 2I–L). The upper inner incisors are in contact and bicuspidate (Figure 2I, J). The lower incisors are bicuspidate (Figure 2K, L). The maxillary and mandibular toothrows have conspicuous diastemata between the premolars and molars (Figure 2D, H, M). The molars are not cuspidate (Figure 2M).

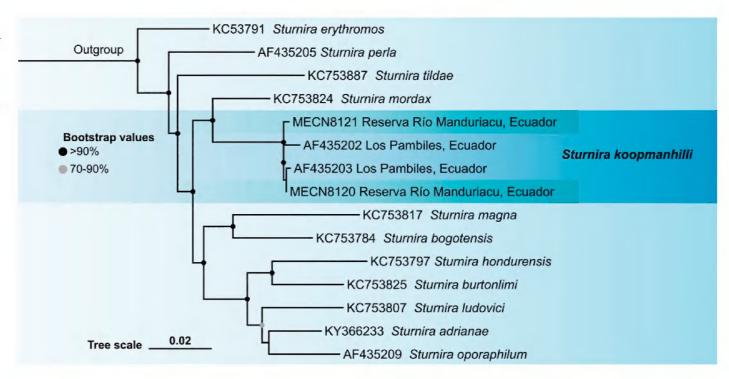
All voucher specimens reviewed were adults with soft, woolly pelage (Figure 3A–D). In very old specimens, the pelage appeared stained orange by shoulder-gland secretions (epaulettes) (Figure 3A–D). For example, QCAZ 4256 and 4258 were brown dorsally and lighter ventrally. Between the shoulders there is four-band color pattern (Figure 3E), while ventrally there is a three- to four-band color pattern (Figure 3F). In living animals, there is a rounded outgrowth, at the base of the spear on top of the narial pad on each side of the nose leaf (Figure 4). Our measurements are consistent with the original species description and the specimens used for comparisons (Table 1).

The 1140-bp-long *cytb* sequences from MECN 8120 and 8121 were part of a multiple sequence alignment (MSA) of 15 DNA sequences, 10 of which represented the full-length of the *cyt-b* gene. Within *Sturnira*, the subgenus *Sturnira* was recovered as monophyletic. Intraspecific relationships between *S. mordax*, *S. hondurensis*, *S. burtonlimi*, and *S. ludovici* are strongly supported (Figure 5), while the support for all other species is considered moderate. Genetic distances between groups exceed 4% and reach up to 9%. Specimens

Table 1. Measurements of so	ome specimens of Sturnira	<i>koopmanhilli</i> examined.
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	Present study									M. C. II	
Measurements	QCAZ 682 ♀	QCAZ 4256 ♂	QCAZ 4258 ♂	QCAZ 18711 💍	MECN 6802 ♂	MECN 6806 ♀	MECN 7432 ♀	MECN 7475 ♂	MECN 8120 ♂	MECN 8121 ♀	McCarthy et al. 2006
Head & body length	86.24	95.45	88.68	83.94	74.13	69.53	94	79	72	72	72–88
Hindfoot length	16.41	15.23	16.76	16.51	17.47	12.7	18	12.23	16	16	11–19
Ear	11.8	_	17.31	17.68	17.8	13.64	19	14	18	20	13–20
Forearm	50.17	53.52	51.83	51.17	52.49	49.79	50.7	48.58	51	47	48.1–52.4
Weight		_	_	_	35	27	32	27.5	34	25	25.5-36

**Figure 5.** Maximum-likelihood phylogenetic tree inferred from the *cytb* analyses of selected *Sturnira* species. The tree includes the samples, MECN 8120 and 8121 from the Reserva Río Manduriacu, Ecuador. Branches show bootstrap maximum-likelihood support (in percent).



MECN 8120 and 8121 are nested within the *S. koopmanhilli* clade and have a genetic distance of 0.45% from other members of the clade, 0.264% between themselves, and 4.13% from *S. mordax*, the closest species to *S. koopmanhilli*. Support for the *S. koopmanhilli* clade was 100% and 68% among its individuals.

## **DISCUSSION**

We extend the distribution of *Sturnira koopmanhilli* to the eastern slopes of the northern Andes. Valuable images and genetic material are now available for other researchers interested in identifying this species.

Vargas et al. (2023) argued that investing more time and resources in curatorial work is more efficient than investing in field expeditions to fill the biodiversity data gap. We consider both investments necessary as researchers analyze the distribution of rare and threatened species in the Neotropics. Conservation efforts must be based on the natural history, ecology, and biogeography of species to design appropriate conservation strategies (IUCN 2012); thus, the collection of baseline information remains crucial. Further studies in the Chocó biogeographic region and in Archidona are needed to understand the ecology of this bat species. In addition, the support found among individuals in the *S. koopmanhilli* clade was 68%, and more individuals are needed to increase the support in future phylogenetic analyses (Velazco and Patterson 2013).

Most of the characters analyzed in this paper are consistent with the original description of *S. koopman-hilli*, with the exception of the ventral bands of the pelage (McCarthy et al. 2006). As pointed out by Jarrín and Kunz (2011), color patterns should not be considered as an informative character for taxonomic identification due to the high intraspecific and age variation observed among individuals of the same species. This natural intraspecific variation has been reported in other species such as *S. bakeri* and *S. lilium* (Peterson and Tamsitt 1968; Rodríguez-Segovia 2022). As such, we recommend the use of a combination of diagnostic anatomical characters to identify bat species.

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# **ADDITIONAL INFORMATION**

#### Conflict of interest

The authors declare that no competing interests exist.

#### **Ethical statement**

No ethical statement is reported.

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## **Author contributions**

Conceptualization: MARS. Data curation: MARS. Formal analysis: MARS, JB, LS. Funding acquisition: JB. Investigation: MARS, JB. Methodology: MARS, JB. Visualization: MARS. Validation: MARS. Writing — original draft: MARS. Writing — review and editing: MARS, JB, LS.

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#### Data availability

All the data that support the findings of this study are available in the main text.

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## **APPENDICES**

### **Appendix 1.** Localities for *Sturnira koopmanhilli*.

Museums and Institutions: CM = Section of Mammals, Carnegie Museum of Natural History, Pittsburgh, Pennsylvania; EPN (currently MEPN) = Instituto de Ciencias Biológicas, Escuela Politécnica Nacional, Quito, Ecuador; UV = Departamento de Biología, Universidad del Valle, Cali, Colombia; MECN = División de Mastozoología, Instituto Nacional de Biodiversidad, Quito, Ecuador; QCAZ = Museo de Zoología QCAZ, Pontificia Universidad Católica del Ecuador, Quito, Ecuador; MUA = Colección Teriológica de la Universidad de Antioquia (CTUA), Departamento de Antioquia, Colombia; CSJ-M = Museo de Ciencias de la Salle, Bogotá, Colombia.

ECUADOR — CHIMBORAZO • Pallatanga (locality #9, Figure 1); 02°00′00″S, 078°57′00″W; 1650 m alt.; 27 Feb. 1967; M. Olalla; 1 Q, EPN 6722/MEPN 2569 — ESMERALDAS • Reserva Ecológica Cotacachi-Cayapas, confluence of unnamed river with Río las Piedras, Los Pambiles (type locality, locality #8, Figure 1); 00°32′00″N, 078°37′00″W; 1200 m alt.; 23–26 Jul 1985; P. Mena V. and J. Regalado B; 5 ♂ and 6 Q, EPN 2269, 2270, 2276, 2279–2281, 2283, 2284, 2287–2289; 23–27 Nov. 1991; I. Manzano and T.J. McCarthy; 9 ♂ and 12 Q, CM 112804–112821, EPN 9770 (holotype), 9771, 9772 — PICHINCHA • Quito, Pacto, Mashpi Lodge, Seven Layers Trail (locality #13, Figure 1); 00°09′59″N, 078°52′59″W; 931 m alt.; 7 May 2021; Marco Rodríguez; 1 Q, MECN 6806, skin and skull • Quito, Pacto, Mashpi Lodge, Luminous Trail (locality #14, Figure 1); 00°10′00.3″N, 078°52′37″W, 954 m alt.; 10 Mar. 2021; Marco Rodríguez; 1 ♂, MECN 6802, skin and skull • Río Pachijal, Comunidad Las Tolas (locality #10, Figure 1); 00°12′00″N, 078°58′00″W; 600 m alt.; no date; R. Arcos y P. Moreno; 1 unknown sex, EPN 6095.

COLOMBIA — DEPARTAMENTO DE ANTIQUIA • Municipio de Urrao, Vereda Calles, en un bosque húmedo ubicado en un pequeño afluente del Río Calles, el cual desemboca en el Río Jegamecoda, hacia el suroccidente del Parque Nacional Natural, Las Orquídeas (locality #11, Figure 1); 06°31′00″N, 076°17′00″W; 1350 m alt.; 13–14 Jul. 1986; Hermano Lasallista Marco Antonio Serna; 1 ♂ and 1 ♀, CSJ-M 529 and CSJ-M 230 • Municipio de Amalfi, Vereda La Cancana (locality #12, Figure 1); 06°47′00″N, 075°06′00″W; 1500 m alt.; 5 and 7 Jan 1988; Hermano Lasallista Marco Antonio Serna; 2 🖒, CSJ-M60 and MUA 10641 — **DEPARTAMENTO DE CHOCÓ •** 4 km N La Italia (locality #1); 04°56′45″N, 076°25′08″W; 300 m alt.; 6 Sep. 1988; M.S. Alberico; 1 ♀, UV 10019 • Chocó, Alto de Galapagos, near San José del Palmar (locality #2, Figure 1); 04°52′47″N, 076°11′01″W, 2000 m alt.; 14 May 1985; J. Albornoz R. and L.A. Neira; 3 ♀, UV 4440–4442 • Chocó, Alto de Oso, 8 km W La Italia (locality #3, Figure 1);  $04^{\circ}52'10''N$ ,  $076^{\circ}11'01''W$ ; 2000 m alt.; 2 Aug. 1986; M. S. Alberico;  $1 \frac{1}{2}$  and  $1 \frac{1}{2}$ , UV 7443, 7445 — **DEPAR-**TAMENTO DEL VALLE DEL CAUCA • Campamento del Río Azul (locality #4, Figure 1); 03°56′24″N, 076°45′00″W; 850 m alt.; 24–25 Jul. 1993; V. Rojas D; 4 &, UV 11556, 11566, 11567, 11569 • Valle del Cauca, Campamento del Río Azul, confluence of Río Azul at Río Calima (locality #5, Figure 1), 03°55′00″N, 076°42′00″W; 560 m alt.; 18 Apr. 1993; V. Rojas D; 1♂ and 1♀, UV 11190 and UV 11198 • Valle del Cauca, Quebrada de la Delfina, Vereda Aguas Lindas (locality #6, Figure 1); 03°49′44″N, 076°14′11″W; 1200 m alt.; 23 Feb. 1998; C.A. Saavedra; 3 ♂, UV 11724–26 — DEPARTAMENTO DE NARIÑO • Reserva Natural "La Planada" (locality #7, Figure 1); 01°07′00"N, 077°52′48"W; 1950 m alt.; 29 Jul. 1981; M.S. Alberico; 1 3, UV 2926.

#### **Appendix 2.** Sturnira ludovici used for this research.

ECUADOR — **CARCHI** • Mira, Jacinto Jijón y Camaño, La Florida, el Cielito; 00°46′00.9″N, 78°15′12.2″W; 972 m alt.; 18 Mar 2013; Carlos Boada, 2 ♂, QCAZ 14739 and QCAZ 14740 • Tulcán, La Centella; 00°48′51″N, 078°00′53″W; 2600–2800 m alt.; 30 Jan 2013; Carlos Boada; 1♂, QCAZ 14738.